

## RESEARCH ARTICLE

### OPTIMIZATION OF NATURAL POLYMERIC ENTERAL FEED FORMULA USING RESPONSE SURFACE METHODOLOGY

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The cereal mix, soy flour, germinated green gram flour, papaya powder, chekkurmani leaves powder, tapioca flour and spice mix were combined to develop natural polymeric balanced enteral feed. The level of incorporation of tapioca flour (20%), chekkurmani leaves powder (10%), papaya powder (10%), and spice mix (5%) was kept constant. The mixing level of cereal mix ( $X_1$ ), soy flour ( $X_2$ ), and germinated green gram flour ( $X_3$ ) was determined by Central Composite Rotatable Design (CCRD) of Response Surface Methodology (RSM) with five level of each factor. The analyzed responses were total carbohydrate (g %), protein (g %), fat (g %), fiber (g %), energy (Kcal), sensory profile, bulk density (g/ml) and water absorption capacity (%). Cereal mix had positive influence on carbohydrate, grainy nature, viscosity, bulk density and water absorption capacity; Dehusked soy flour had positive influence on protein, fat, fiber, energy and viscosity; and germinated green gram flour had positive influence on fiber, sensory acceptability score, starchy mouth coating, stickiness and grainy nature of the developed feed. Thus the 42g of cereal mix, 10.9g of dehusked soy flour and 4g of germinated green gram flour could be mixed with maximum desirability of 52.6% for the production of balanced polymeric enteral feed.

**Key words:** Polymeric enteral feed, Optimization, Responses, Central Composite Rotatable Design (CCRD), Desirability.

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## INTRODUCTION

Enteral nutrition formulas are used as nutritional replacements for patients who are unable to get enough nutrients in their diet. These formulas are taken by mouth or through a feeding tube and are used by the body for energy and to form substance needed for normal body functions. Patients with acquired immune deficiency syndrome (AIDS), cancer, burns, infections, prolonged kidney, liver, lung, pancreatic and stomach problems, surgery, trauma, vomiting and prolonged diarrhoea may be more likely to need enteral feeding (Vishwanath, 2003). Many different polymeric commercial formulas are available with the protein constituting 12 to 20 %, carbohydrates 40 to 60 % and fats 30 to 40 % of total calories. In the standard formulas, the ratio of nitrogen to non-protein calories is about 1 g/150 kcals. In the high-nitrogen formulas, this ratio can be as high as 1 g/75 kcals. The amount of fiber ranges between 6 and 14 g/1000 kcals. Blenderized natural foods are available commercially or can be prepared by the patient. The commercial blenderized food formulas are prepared from milk, beef, fruits, vegetables and fiber; hence their nutrient content is not determined precisely, and their nutritional completeness is not ensured. Commercial blenderized food products are usually more expensive than polymeric formulas. Patients who use enteral feeding in the home can prepare blenderized foods from regular foods in the household.

If this practice is used, the nutritional adequacy of the blenderized foods must be ensured, particularly if patients are receiving long-term therapy (Shils *et al.*, 2006). The administration of enteral feed provides effects that are far beyond those of merely administering macro and micro nutrients. Rather the processing of nutrients via the GI tract stimulates a complex response that has implications for body composition and for immunologic integrity. Route and type of nutrition are important aspects of successful patient recovery (Kudsk, 2007).

Foods with a specific targeted health effect have become a matter of great attention among the researchers, medical practitioners, food companies and marketing agencies, because the demand for such product is enormous and growing fast day-by-day. With this view the present study was planned to formulate a natural polymeric enteral feed, to study the effect of major ingredients (cereal mix, dehusked soy flour and germinated green gram flour) on quality of the feed, and to optimize the level of ingredients for nutritionally balanced feed using response surface methodology.

## MATERIALS AND METHODS

### Selection of ingredients

The ingredients such as rice (*Oryza sativa*), proso millet (*Panicum miliaceum*), green gram (*Vigna radiate*), tapioca flour (*Manihot esculenta*), papaya (*Carica papaya*), soybean (*Glycine max*), fenugreek seeds (*Trigonella foenum-graecum*), cumin seeds (*Cuminum cyminum*), coriander seeds (*Coriandrum sativum*) and cardamom (*Amomum subulatum*) were procured from the local

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market in Salem District; wheat germ (*Triticum aestivum*) was obtained from Shakthi Murugan Argo Foods, Avinashi and Chekkurmani leaves (*Sauropus androgynus*) from Fruit farm, Karumandurai; Horticulture Department of Agriculture Institute, Virudhachalam and Tamil Nadu Agricultural University, Coimbatore. All the ingredients were selected by considering its availability, nutritional and health benefits.

#### Processing of ingredients

The selected ingredients were processed by adopting optimum condition of processing specific to each ingredient. The parboiled milled rice was roasted at 80°C for 20 minutes; proso millet was soaked in cold water for 24 hours and sun dried for 6 hours; thin sliced papaya was dried in cabinet tray drier at 70-80°C for 48 hours; soybean was soaked in cold water for 24 hours, dehusked, steamed for 20 minutes and sundried for 2 days; the fresh Chekkurmani leaves were shade dried at 30.5°C for 48 hours; the green gram soaked in cold water for 12 hours was germinated in sprout maker for 12 hours and sun dried for 2 days; the spices such as fenugreek seeds, cumin seeds, coriander seeds and cardamom was roasted till it becomes brown at 79°C for 15 minutes. All the processed ingredients were powdered and stored in air tight container. The processing yield and the changes in proximate composition on processing of ingredients were determined using standard procedures.

#### Preparation of enteral feed

The natural polymeric enteral feed was prepared by mixing cereal mix, dehusked soy flour and germinated green gram flour at 55%; tapioca flour at 20%; papaya powder at 10%; chekkurmani leaves powder at 10%; and spice mix at 5%. The proportion of each ingredient was calculated on the basis of recommended level of each food group for the preparation of low cost balanced diet for Indian adult man/women with sedentary activity (Gopalan *et al.*, 2008). The combination of cereal mix were selected in such a way that it could able to provide 63g of carbohydrate (55% of calories), 18.7g of protein (15% of calories) and 19.4g of fat (30% of calories). The spice mix was prepared by combining the selected spices in equal proportion.

#### Experimental design for optimization

The Central Composite Rotatable Design (CCRD) for three variables at five levels (Table 1) was used to study the response pattern and to determine the optimum combination of variables such as cereal mix ( $X_1$ ), dehusked soy flour ( $X_2$ ) and germinated green gram flour ( $X_3$ ) (Yadav and Sharma, 2008).

#### Responses for optimization

The experimental variables of 20 runs (Table 1) were analyzed for its carbohydrate, protein, fat, fiber, (Sadasivam and Manickam, 2005); energy (Gopalan *et al.*, 2008); bulk density (Wang and Kinsella, 1976); water absorption capacity (Janicki and Walczak, 1960) and sensory profile (Bhat and Sharma, 1989). The sensory profile of the formulated enteral feed was done by boiling 10g of the feed formula from each experimental run with 100ml of water. A second order polynomial regression equation was fitted to the data of all responses.

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \beta_{11} X_1 X_1 + \beta_{22} X_2 X_2 + \beta_{33} X_3 X_3$$

Where  $\beta_0$ ,  $\beta_1$ ,  $\beta_2$ ,  $\beta_3$ ,  $\beta_{11}$ ,  $\beta_{22}$ ,  $\beta_{33}$ ,  $\beta_{12}$ ,  $\beta_{13}$ ,  $\beta_{23}$  were the regression coefficients,  $X_1$ ,  $X_2$ ,  $X_3$ , were the independent variables and Y was the dependent variable.

## RESULTS AND DISCUSSION

#### Processing yield

The processing yield of wheat germ and the selected spices were in the range of 91-94%; cereals in the range of 83-88%; germinated green gram at 88%; soy bean at 71%; papaya at 8.8% and chekkurmani leaves at 29.7%.

#### Nutritional composition of processed ingredients

The changes in nutritional composition of the selected ingredients on processing (Table 2) reveal that there was a significant ( $p < 0.05$ ) increase in carbohydrate, protein, fat and fiber content of all selected ingredients on processing, where as the moisture content of all ingredients; ash content of green gram, soy bean and fenugreek seeds were decreased significantly ( $p < 0.05$ ).

#### Estimated response levels of experimental variables

The estimated response levels of experimental variables revealed that the carbohydrate content was ranged from 62.037 to 65.199 g%; protein from 17.32 to 19.9 g%; fat from 4.18 to 5.17 g%; fiber from 3.93 to 4.12 g%; energy from 364.99 to 371.438 kcal; sensory acceptability score from 14.85 to 16.3; starchy mouth coating score from 1.2 to 1.7; grainy nature score from 1.3 to 1.75; stickiness score from 1.25 to 1.65; viscosity score from 1.35 to 1.75; water absorption capacity score from 179 to 242.5% and bulk density from 0.625 to 0.75 g/ml. Mepba *et al* (2007) reported that the low bulk density could be an advantage in the formulation of baby food and enteral formulas where high nutrient to low bulk density is desired.

#### Influence of variables

The regression analysis of the experimental data (Table 3) showed that the coefficient of determination,  $R^2$  was above 80% in responses such as carbohydrate, protein, fat and fiber content of the enteral feed (significant at  $p < 0.01$ ); 57.14% in stickiness (significant at  $p < 0.05$ ) and 51.91% in overall acceptability score (significant at  $p < 0.05$ ). The other responses were not much influenced for its variation by the independent variables. As per the calculated F-value, all the models except energy, starchy mouth coating, grainy nature, viscosity, bulk density and water absorption capacity were considered adequate for predicting the responses and interpreting the effect of independent variables. The sign and magnitude of the coefficients indicated the effect of the variables on the responses. Negative sign of a coefficient at linear level indicated decrease in response value with an increase in the level of variables. The incorporation of cereal mix significantly ( $p < 0.05$ ) increased the carbohydrate content and decreased the protein and fiber content as well as the level of stickiness of the feed. The protein, fat, crude fiber and energy content of the feed was improved significantly by the incorporation of dehusked soy flour ( $p < 0.05$ ). At the same time, the carbohydrate content and stickiness of the feed was reduced by increasing the level of dehusked soy flour in the feed (significant at  $p < 0.05$ ). Soy protein plays an important role in many food products because of their nutritional value, improving functional properties (water and fat absorption, emulsion, whipping) and for increasing total protein content and improving the essential amino acid profile (Lusas and Riaz, 1995). The

**Table 1. Coded variables for the experimental design**

A. Levels of independent variables						
Variables	Code	Coded level				
		-2	-1	0	+1	+2
Cereal mix	X <sub>1</sub>	36	38	40	42	44
Dehusked Soy flour	X <sub>2</sub>	6	8	10	12	14
Germinated green gram flour	X <sub>3</sub>	3	4	5	6	7

B. Experimental plan				
X <sub>1</sub>	X <sub>2</sub>	X <sub>3</sub>	Number of experiments	
±1	±1	±1	8	
±2	0	0	2	
0	±2	0	2	
0	0	±2	2	
0	0	0	6	

**Table 2. Nutritional compositions of processed ingredients in g %.**

Ingredients	Moisture	Protein	Carbohydrate	Fat	Fiber	Ash
Rice	0.15±0.07 <sup>b</sup>	6.3±0.469	88.17±0.17 <sup>a</sup>	0.25±0.07	0.305±0.007	5.31±0.014 <sup>a</sup>
Wheat germ	0.75±0.07 <sup>b</sup>	29.67±0.09	58.3±0.258 <sup>a</sup>	6.25±0.35	1.51±0.01	5.52±0.228 <sup>a</sup>
Prosomillet	0.85±0.07 <sup>b</sup>	14.2±0.095	80.47±0.275 <sup>a</sup>	1.25±0.07	2.505±0.007	1.55±0.07
Germinated Green gram	2.5±0.07 <sup>b</sup>	27.45±0.46 <sup>a</sup>	64.375±0.298 <sup>a</sup>	1.35±0.07	4.615±0.02	2.05±0.07 <sup>b</sup>
Tapioca	9.1±0.14 <sup>b</sup>	1.75±0.70 <sup>a</sup>	76.425±0.377 <sup>a</sup>	0.405±0.007	2.205±0.007 <sup>a</sup>	3.1±0.14 <sup>a</sup>
Papaya	4.33±0.042 <sup>b</sup>	7.22±0.17 <sup>a</sup>	75.5±0.583 <sup>a</sup>	1.1±0.14 <sup>a</sup>	10.2±0.282 <sup>a</sup>	3.1±0.14 <sup>a</sup>
chekkurmaani leaves	1.45±0.07 <sup>b</sup>	26.9±1.98 <sup>a</sup>	38.075±0.095 <sup>a</sup>	10.2±0.28 <sup>a</sup>	4.5±0.14 <sup>a</sup>	11.05±0.07 <sup>a</sup>
Dehusked Soy bean	1.55±0.07 <sup>b</sup>	48.11±1.43 <sup>a</sup>	24.07±0.095 <sup>a</sup>	18.25±0.35	4.45±2.12	1.775±0.035 <sup>b</sup>
Fenugreek Seeds	1.25±0.07 <sup>b</sup>	26.45±0.19	54.125±0.095 <sup>a</sup>	6.35±0.07	7.7±0.141	2.505±0.007
Cumin seeds	1.85±0.07 <sup>b</sup>	25.34±0.72 <sup>a</sup>	43.07±0.095 <sup>a</sup>	10.2±0.282 <sup>b</sup>	9.3±0.424 <sup>b</sup>	8.75±0.014 <sup>a</sup>
Coriander Seeds	0.85±0.07 <sup>b</sup>	14.625±0.17	27.15±0.173 <sup>a</sup>	15.15±0.21	36.95±0.07 <sup>a</sup>	6.1±0.141 <sup>a</sup>
Cardamom	3.65±0.07 <sup>b</sup>	13.4±0.216 <sup>a</sup>	52.075±0.095 <sup>a</sup>	3.4±0.565	23.9±0.14	6.15±0.212 <sup>a</sup>

<sup>a</sup>- significant increase at p<0.05; <sup>b</sup>- significant decrease at p<0.05 in comparison with raw ingredients indicated in nutritive value of indian foods by Gopalan, 2008.

**Table 3. Regression coefficients for the response variables**

Coefficients	Carbohydrate	Protein	Fat	Fiber	Energy	Sensory acceptability score	Starchy mouth coating	Stickiness	Grainy nature	Viscosity	Bulk density	WAC
β <sub>0</sub>	63.40	18.28	4.62	4.01	368.34	15.19	1.53	1.48	1.45	1.61	0.68	192.01
β <sub>1</sub>	0.22 <sup>**</sup>	-0.13 <sup>*</sup>	-0.056 <sup>ns</sup>	-0.019 <sup>*</sup>	-0.17 <sup>ns</sup>	-0.092 <sup>ns</sup>	0.024 <sup>ns</sup>	-0.043 <sup>**</sup>	0.047 <sup>ns</sup>	-0.013 <sup>ns</sup>	0.000 <sup>ns</sup>	1.82 <sup>ns</sup>
β <sub>2</sub>	-0.85 <sup>*</sup>	0.51 <sup>*</sup>	0.27 <sup>*</sup>	0.052 <sup>*</sup>	1.02 <sup>**</sup>	-0.063 <sup>ns</sup>	0.025 <sup>ns</sup>	-0.059 <sup>**</sup>	-0.042 <sup>ns</sup>	0.021 <sup>ns</sup>	0.000 <sup>ns</sup>	-2.29 <sup>ns</sup>
β <sub>3</sub>	0.011 <sup>ns</sup>	0.026 <sup>ns</sup>	-0.048 <sup>ns</sup>	0.026 <sup>*</sup>	-0.28 <sup>ns</sup>	0.046 <sup>ns</sup>	0.062 <sup>ns</sup>	0.012 <sup>ns</sup>	0.018 <sup>ns</sup>	0.029 <sup>ns</sup>	0.000 <sup>ns</sup>	-1.52 <sup>ns</sup>
β <sub>12</sub>	0.012 <sup>ns</sup>	-0.010 <sup>ns</sup>	-0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	-0.031 <sup>ns</sup>	0.19 <sup>ns</sup>	-0.013 <sup>ns</sup>	0.000 <sup>ns</sup>	0.025 <sup>ns</sup>	-0.019 <sup>ns</sup>	0.000 <sup>ns</sup>	-4.88 <sup>ns</sup>
β <sub>13</sub>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	0.026 <sup>ns</sup>	0.044 <sup>ns</sup>	0.062 <sup>ns</sup>	-0.056 <sup>**</sup>	-0.13 <sup>ns</sup>	0.000 <sup>ns</sup>	-0.016 <sup>ns</sup>	-4.00 <sup>ns</sup>
β <sub>23</sub>	0.013 <sup>ns</sup>	-0.013 <sup>ns</sup>	-0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	-0.032 <sup>ns</sup>	0.019 <sup>ns</sup>	0.025 <sup>ns</sup>	-0.044 <sup>ns</sup>	-0.13 <sup>ns</sup>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>
β <sub>11</sub>	0.030 <sup>ns</sup>	0.018 <sup>ns</sup>	0.036 <sup>ns</sup>	0.000 <sup>ns</sup>	0.52 <sup>ns</sup>	0.16 <sup>ns</sup>	-0.018 <sup>ns</sup>	0.000 <sup>ns</sup>	0.000 <sup>ns</sup>	-0.025 <sup>ns</sup>	0.012 <sup>ns</sup>	12.71 <sup>**</sup>
β <sub>22</sub>	0.074 <sup>ns</sup>	-0.000 <sup>ns</sup>	0.019 <sup>ns</sup>	0.000 <sup>ns</sup>	0.43 <sup>ns</sup>	0.11 <sup>ns</sup>	-0.018 <sup>ns</sup>	-0.011 <sup>ns</sup>	0.026 <sup>ns</sup>	-0.025 <sup>ns</sup>	-0.010 <sup>ns</sup>	5.90 <sup>ns</sup>
β <sub>33</sub>	0.041 <sup>ns</sup>	0.000 <sup>ns</sup>	0.033 <sup>ns</sup>	0.000 <sup>ns</sup>	0.50 <sup>ns</sup>	0.32 <sup>*</sup>	0.000 <sup>ns</sup>	-0.011 <sup>ns</sup>	0.035 <sup>ns</sup>	-0.051 <sup>ns</sup>	0.000 <sup>ns</sup>	5.11 <sup>ns</sup>
R <sub>2</sub>	88	96.62	81.85	87.52	15.98	51.91	-26.30	57.14	45.93	-21.97	-1.13	32.80
F	17.81 <sup>*</sup>	61.42 <sup>*</sup>	10.52 <sup>*</sup>	15.80 <sup>*</sup>	1.40 <sup>ns</sup>	3.28 <sup>**</sup>	0.56 <sup>ns</sup>	3.81 <sup>**</sup>	2.79 <sup>ns</sup>	0.62 <sup>ns</sup>	1.02 <sup>ns</sup>	2.03 <sup>ns</sup>

\* - Significant at p<0.01; \*\* - Significant at p<0.05; ns - Not significant. WAC - Water Absorption Capacity

incorporation of germinated green gram flour showed a significant (p<0.05) increase in crude fiber content of the feed but did not influence all the other responses significantly. At the interactive level, level of one variable could be increased while that of other decreased to get the same response value. The cereal mix and germinated

green gram flour at interactive level (Fig. 1) affected the stickiness of the feed, negatively significantly at p<0.05.

**Optimized level of ingredients for enteral feed**

The criteria used for optimization along with predicted values of responses have been presented in Table 4. As

Table 4. Criteria for optimization and predicted response values

Constraints	Goal	Lower Limit	Upper Limit	Importance	predicted value
Carbohydrate (g %)	Maximize	62.04	65.20	5	63.31
Protein (g %)	Maximize	17.32	19.19	5	18.37
Fat (g %)	Minimize	4.18	5.17	5	4.81
Fiber (g %)	Maximize	3.93	4.12	5	4.003
Energy (kcal %)	Maximize	364.99	371.44	5	369.97
Sensory acceptability score	Maximize	14.85	16.3	5	15.56
Starchy mouth coating	Minimize	1.2	1.7	5	1.40
Stickiness	Minimize	1.25	1.65	5	1.45
Grainy nature	Minimize	1.3	1.75	5	1.57
Viscosity	Minimize	1.35	1.75	5	1.47
Bulk density	Maximize	0.625	0.75	5	0.726
Water absorption capacity	Maximize	179	242.5	5	215.14

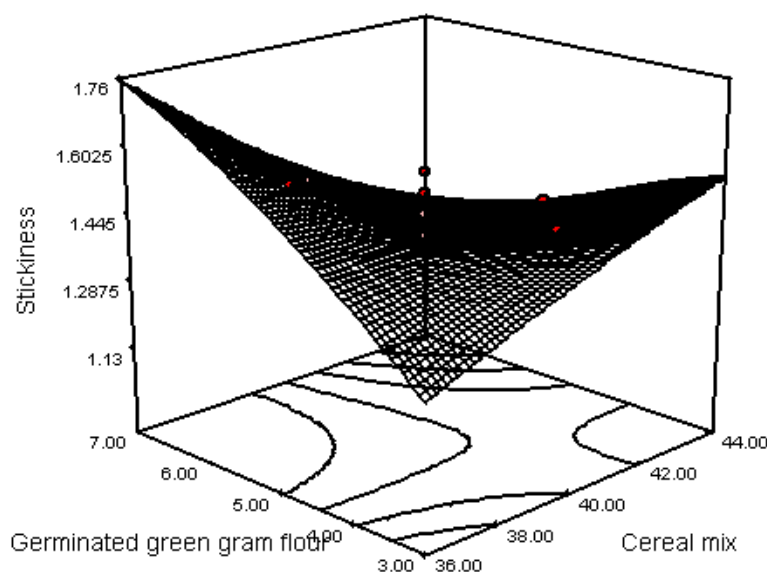


Fig. 1 3D Response surface graph showing the effect of cereal mix and germinated green gram flour on stickiness of enteral feed

per the goal set for each responses in numerical optimization process of response surface methodology, 32 solutions with minimum desirability of 45.7% was resulted. The suggested solution with the maximum desirability of 52.6% indicated the mixing level of cereal mix, dehulled soy flour and germinated green gram flour at 42g, 10.9g, and 4g respectively.

#### Conclusion

The dehulled soy flour plays a dominant role in improving the quality of the enteral feed on the basis of determined responses. The incorporation of chekkurmani leaves powder imparted green colour to the formulated enteral feed. The compromise optimum level of independent variables with maximum desirability of 52.6% may be recommended for preparing a good quality polymeric balanced enteral feed formula.

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